APPENDIX

AMENDMENTS TO THE SPECIFICATION

Please amend the paragraph from page 7, line 1, to page 9, line 3, as follows:

Surprisingly, it has now been found that an immunogenically active component which comprises inactivated *Sarcocystis neurona* cells or antigens, subunit proteins or plasmid DNA; inactivated *Neospora hughesi* cells or antigens, subunit proteins or plasmid DNA; or mixtures thereof may be administered in the form of a vaccine composition to prevent or ameliorate EPM disease in equines, particularly horses. Antigens derived from *Sarcocystis neurona* or *Neospora hughesi* may be obtained using conventional procedures such as outer membrane extraction. Plasmid DNA derived from *Sarcocystis neurona* or *Neospora hughesi* may be obtained via isolation from sources such as the fluids or tissues of equine mammalians diagnosed to have EPM. Such sources include cerebral spinal fluid or sections of spinal cord or brain. Alternatively, the precursor of the infectious stage in horses (sporocyst or cyst) may be obtained from feces or intestinal scrapings of opossums or other wild life present in endemic locales. *Sarcocystis* Spp. or *Neospora* SPP. cells, thus obtained, may be maintained in the infected equine or in suitable tissue culture media, such as RPMI 1640 medium or in cells known in the art such as African green monkey kidney (Vero) cells or equine dermal (E. Derm) cells. The *Sarcocystis* Spp. or *Neospora* Spp. protozoa may then be separated from the tissue culture of cell media using conventional techniques such as centrifugation, filtration, or the like. A useful starting isolate for the vaccines of the invention include, for example, for *Sarcocystis neurona*, the isolate designated SN3; other such isolates are those known as SN1, SN2, SN4, SN5, SN6, UCD-1, UCD-2 and UCD-3 and are variously available from the University of Kentucky, Dr. J.P Dubey at the USDA, U. of California – Davis, Oregon State University, the University of Missouri and others. A culture of one such *Sarcocystis neurona* isolate designated SNg, originally isolated from the intestinal scrapings of the opossum and confirmed to be a representative *Sarcocystis neurona* by PCR, was deposited ~~with the ATCC~~ on January 25, 2001 in the American Type Culture Collection (ATCC), 10801 University Boulevard, Manassas, Virginia 20110-2209, U.S.A., and given ATCC Accession No. PTA-2972. The deposit was made under the conditions mandated by 37 C.F.R. § 1.808 and is being

maintained pursuant to the Budapest Treaty. A useful starting isolate for the vaccines of the invention include, for example, for *Neospora hughesi*, the isolate designated NEQ1; another such isolate is that known as NE1, which has been described by Antoinette Marsh *et al*, Journal of Parasitology, 84 (5), 1998, pp 983-991. A culture of one such Neospora hughesi isolate has been deposited with the ATCC and given ATCC Accession No. 209622 (NE1) as disclosed in US 6,071,737. Surprisingly, it has now been found that protozoan parasites such as *Sarcocystis* spp. or *Neospora* Spp. may be propagated in increased yield and increased active viability via cell culture propagation by growing suitable cells to a monolayer having a confluency of about 80%-100% in a growth media; decanting the growth media; refeeding the cells with fresh growth media; inoculating the cells with merozoites or tachyzoites; after 4-12 days, decanting the growth media; and refeeding the inoculated cells a second time with growth media. Cells suitable for use in the method of the invention include cells such as E. Derm cells, Vero cells, Maiden Darby Bovine Kidney (MDBK) cells, Canine Monocyte (DH82) cells, Mouse Monocyte (P388) cells, Fetal Rhesus Monkey Kidney cells, Feline Kidney (FKCU) cells, Maiden Darby Canine Kidney (MDCK) cells, Baby Hamster Kidney (BHK21) cells, or the like, preferably E. Derm or Vero cells, more preferably E. Derm cells.